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GDF11 exerts anti-inflammatory activity in the mouse model of acute liver injury

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INTRODUCTION

Drug caused hepatotoxicity, viral hepatitis, alcohol consumption, and other gastrointestinal tract comorbidities contribute to liver injury. Current scientific data prove that Transforming growth factor β (TGFβ) superfamily plays a significant role in hepatitis development. It is confirmed that Growth differentiation factor 11 (GDF11), the member of TGFβ superfamily affects liver pathologies. The positive effect of GDF11 is confirmed in cirrhosis, liver fibrosis and fatty liver disease. GDF11 is presumed to have anti-inflammatory effects, however, some reports have stated that GDF11 aggravate liver disease.

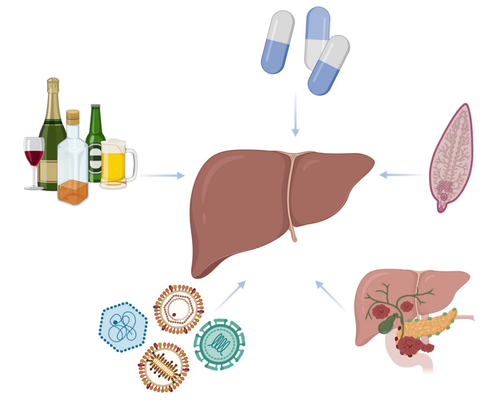

Fig. 1 Major factors affecting liver injury development

The aim of the study was to validate of the acute hepatitis model in mice.
Assessment of GDF11 role in acute liver injury.

METHODS

Male C57BL/6 mice were obtained from the Animal Facility of the University of Lodz, Lodz, Poland. All procedures on animals were approved by the Local Ethical Committee for Animal Experiments (38/ŁB/212/2021). Animals were maintained under a 12 hours light/dark cycle and a constant temperature (22°C) with free access to chow pellets and tap water. D-galactosamine (GalN) was administrated at the dose of 0.7 mg/kg *i.p.*, 30 minutes before TNFα *i.p.* injection (0.1 μg or 0.3 μg/mouse). After 6 hours animals were sacrificed. GDF11 was administrated at the dose of 0.1 mg/kg *i.p.* 15 minutes prior to the GalN injection. Liver samples were collected for histology, RNA and protein isolation. The expression of *Gdf11*, *Tnfα*, *Nos2*, *Bax*, *Il6*, *Il1b, Ptgs2* at mRNA level was assessed by real-time RT-PCR. The expression of SMAD 2/3, TNFα and β-ACTIN was obtained by western blot. ΔΔCt method was applied for calculation of PCR results. Normalization to housekeeping protein was applied for determination of relative protein expression. The hepatitis scoring system was applied for histology assessment. Data present mean ± Standard Error of Mean (SEM). Statistical analysis was performed using GraphPad Software.

RESULTS

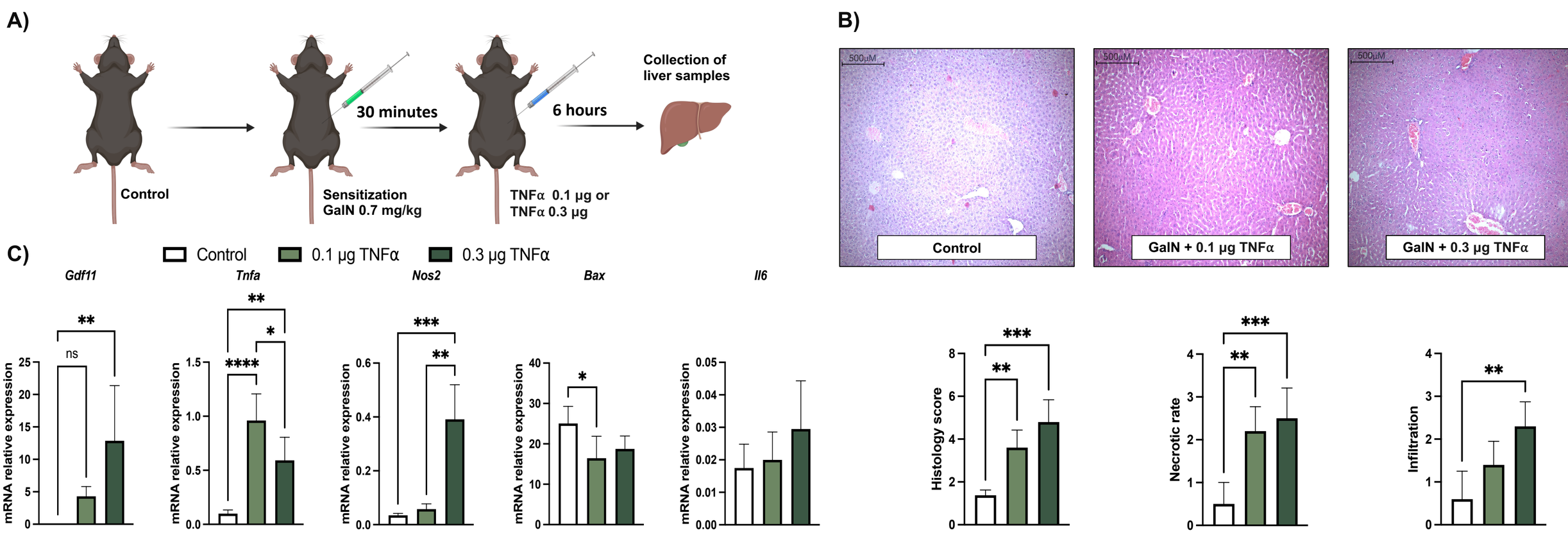

Fig. 2 Experimental design of acute liver injury validation (A). Representative microscopy pictures of a healthy mouse liver and acute liver injury, original magnification ×100, histological score of liver changes (B). mRNA expression of *Gdf11*, *Tnfα*, *Nos2*, *Bax, Il6* in the liver (C). Statistical significance confirmed with one-way ANOVA with post-hoc test * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$, ****$p < 0.0001$.

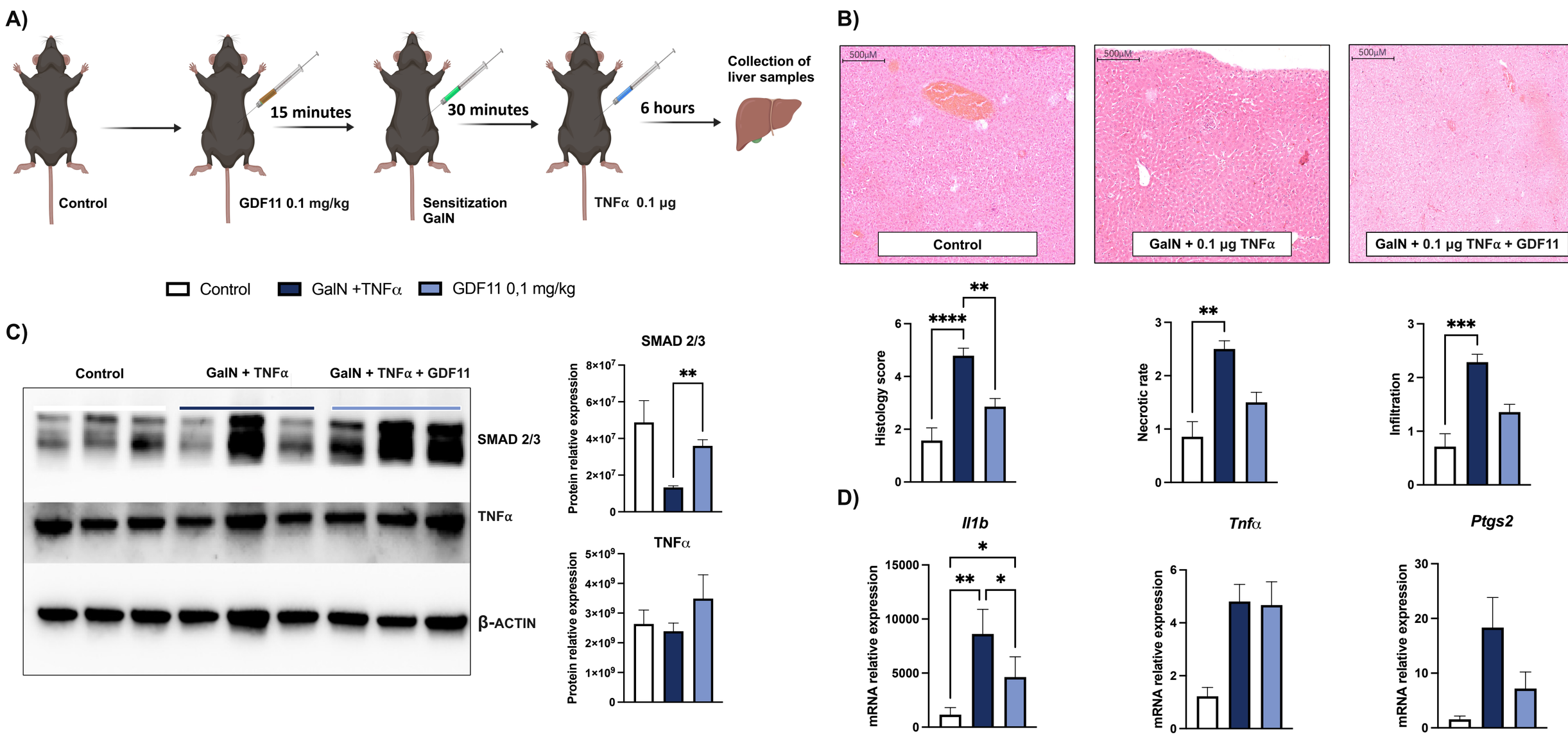

Fig. 3 Experimental design of acute liver injury treated with GDF11 (A). Representative microscopy pictures of healthy mouse liver, acute liver injury and acute liver injury with GDF11, original magnification ×100, histological score of liver changes (B). Western blot and protein relative expression of SMAD 2/3, TNFα and β-ACTIN (C). *Il1b*, *Tnfα*, *Ptgs2* mRNA expression in the liver (D). Statistical significance confirmed with one-way ANOVA and followed with post-hoc test * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$,****$p < 0.0001$.

CONCLUSION

The model of TNFα induced liver injury was validated. The expression of *Gdf11* is altered during the course of acute hepatitis.

GDF11 might be a promising agent against liver injury through canonical signaling pathway regulation.